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Article

Certified Matrix-matched Sulfide Reference Materials for Direct Fe, Cu, and Zn Isotopic Analysis of Simple-matrix Minerals

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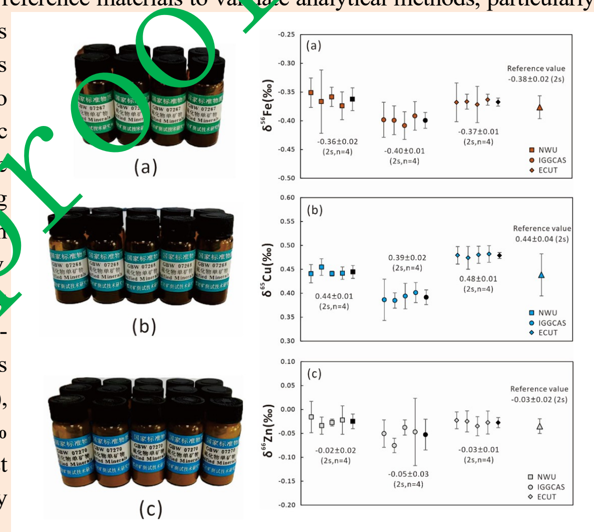
ABSTRACT: Stable isotope systems of iron, copper, and zinc have emerged as powerful tracers in understanding metal sources, migration, and deposit formation processes. Accurate and precise determination of Fe, Cu, and Zn isotopic compositions in sulfide minerals, especially in simple-matrix minerals characterized by their relatively pure composition, with low content of impurity elements and few interfering components, requires matrix-matched reference materials to validate analytical methods, particularly for direct analysis protocols without column chromatography. This study introduces a suite of novel secondary reference materials (NWU-Fe, NWU-Cu, and NWU-Zn sulfide powders) developed to address the critical gap in calibration standards for direct isotopic analysis without column chemistry of simple-matrix minerals. These sulfide powders exhibit excellent homogeneity and stability, fulfilling the requirements for high-precision determination of Fe, Cu, and Zn isotope ratios using MC-ICP-MS without column chemistry. Reference values were derived from interlaboratory comparison across three independent laboratories. The isotopic compositions (δ-values), reported in per mil notation relative to international standards (IRMM-014 for Fe, NIST SRM-976 for Cu, and JMC Lyon for Zn), are as follows: $\delta^{56}Fe = -0.38 \pm 0.02$‰ (2s), $\delta^{65}Cu = 0.44 \pm 0.04$‰ (2s), $\delta^{66}Zn = -0.04 \pm 0.02$‰ (2s). This study provides a robust calibration framework for Fe-Cu-Zn isotopic studies in geochemistry and environmental science.



INTRODUCTION

Significant advances in multi-collector inductively coupled plasma mass spectrometry (MC-ICP-MS) and high-precision analyses of stable Fe, Cu and Zn isotopes have been widely used in mineral deposits[1-4], earth-surface processes[5,6], cosmochemistry[7], palaeoceanography[8], archaeology[9], and medical science[10, 11]. High-precision and high-accuracy measurements are essential for the application of Fe, Cu, and Zn isotopes as geochemical proxies, given the limited isotopic fractionation in natural samples. In traditional analytical methods, column chromatography is a prerequisite for the purification of Fe, Cu, and Zn fractions from complex geological samples, effectively minimizing potential matrix effects and isobaric interferences. However, this process is labor-intensive and time-consuming, often involving the use of hazardous chemicals and generating significant waste. In previous studies, the purification of Fe-Cu-Zn fractions through column chromatography with a high yield used different cation-exchange resins, acids, and columns[12-18]. Nevertheless, the determination of Fe-Cu-Zn isotopic compositions has traditionally relied on bulk rock or bulk mineral concentrate analyses following separation and purification by column chromatography, a complicated and labor-demanding process due to the presence of complex matrix elements.

Recently, *in situ* Fe, Cu, Zn isotope analysis of Fe, Cu, Zn-dominated minerals have been developed by using laser ablation (LA-) combined with MC-ICP-MS, suggesting Fe-Cu-Zn isotopes with good accuracy and precision can be obtained without column chromatography[19-28]. Specifically, femtosecond laser (fsLA-) and nanosecond laser (nsLA-) have been used to evaluate matrix effects during Fe isotope analysis, with water addition during fsLA-MC-ICP-MS analysis significantly reducing these effects and enabling precise measurements to the ~0.1‰ level with non-matrix-matching calibration[27]. Similar approaches have been successfully applied to Cu and Zn isotopes, with accurate ratios obtained under wet plasma conditions[20, 24, 28]. These findings indicate that Fe, Cu, and Zn isotope ratios of Fe, Cu, and Zn-dominated minerals could be accurately measured without chemical purification, while water addition simulates a wet plasma condition, which is often more resistant to matrix effects.

Most recently, the direct measurement of Fe, Cu, and Zn isotope ratios in diverse minerals and the associated matrix effects have been thoroughly investigated[29-31]. Reliable data for Fe, Cu, and Zn isotopes can be respectively obtained in simple matrix minerals without chromatographic separation. Zhang *et al*. (2020)[29] used the standard-sample bracketing (SSB) technique with Ga as an internal standard to determine Cu isotope ratios in twelve Cu-dominated minerals. The measured $\delta^{65}Cu_{without-with}$ values range from −0.04‰ to +0.02‰, indicating that accurate and precise Cu isotope ratios in Cu-dominated minerals can be achieved without column chromatography. Fe isotope ratios of 40 iron-dominated material/mineral samples without column chromatography were investigated by Chen *et al*. (2022)[30] using the SSB technique. The $\delta^{56}Fe_{without-with}$ values ranging from -0.06‰ to +0.06‰ indicate that Fe isotope ratios in Fe-dominated minerals can be achieved without column chromatography, due to the simple matrix and the stability of the machine in high-resolution wet plasma condition. Moreover, Nie *et al*. (2022)[31] directly determined Zn isotopes in sphalerite samples and Zn-rich minerals without column separation. The SSB technique with Cu as the internal standard was used to correct the instrumental mass bias, and the results showed that Zn isotopic ratios obtained without column chromatography are consistent with those obtained with column chromatography, and the $\delta^{66}Zn_{without-with}$ and $\delta^{67}Zn_{without-with}$ values range from -0.04 to +0.01‰ and from -0.06‰ to +0.01‰, respectively. These reproducible results demonstrate that, simple-matrix minerals, such as Fe, Cu, and Zn-dominated minerals (e.g., pyrite, chalcopyrite, and sphalerite), are a critical group of minerals that can be analyzed without complex chemical purification protocols. These minerals are characterized by their relatively pure composition, with low content of impurity elements and few interfering components. These minerals are widely distributed in various types of ore deposits and surficial environments, serving as key carriers for metal isotope tracing and playing an irreplaceable role in studies on ore genesis and element migration[29-31]. The simplicity of their matrix is crucial for achieving high-precision isotopic measurements, as it minimizes potential matrix effects and isobaric interferences that are common in more complex geological samples. This simplicity also enables direct isotopic analysis using MC-ICP-MS. The advantages of this method include reduced reagent consumption, lower environmental impact, faster sample throughput, and enhanced laboratory safety.

The advancements in direct determination of Fe-Cu-Zn isotopes have been successfully applied in samples with simple matrices, such as sulfides, oxides and elemental substances, in geological research[32-38]. In traditional column chemistry, reference materials are indispensable for ensuring the accuracy and precision of isotopic measurements by validating analytical methods, especially for complex matrix samples such as bulk rock. Similarly, for simple matrix minerals, reference materials are crucial for validating direct analysis methods and ensuring reliable results. However, matrix-matched reference materials for direct Fe, Cu, and Zn isotope analysis without column chromatography have not been widely reported.

This study focuses on ore minerals with simple matrices, enabling direct isotopic analysis without chemical purification. We developed a method for the rapid and accurate determination of Fe, Cu, and Zn isotope compositions using MC-ICP-MS. To ensure the accuracy and reliability of this method, it is essential to develop ore mineral reference materials that are matrix-matched and have homogeneous isotope compositions. These materials will serve as robust quality-control standards for Fe, Cu, and Zn isotope analysis.

EXPERIMENTAL

Apparatus and reagents. Candidate reference material sulfide powders (pyrite, chalcopyrite and sphalerite) were respectively prepared in a Class 1000 clean room at the State Key Laboratory of Continental Evolution and Early Life (SKLCEEL), Department of Geology, Northwest University, Xi'an, China. A Savillex® DST-1000 acid purification system was used to purify acids, including HCl, HNO_3 and HF. Deionised water with a resistivity of 18.0 MΩ cm^{-1} was obtained from a Milli-Q water purification system (Millipore, Bedford, MA, USA). The Savillex® vials were cleaned in hot (130 ^{o}C) GR grade HNO_3 for 1 day and GR grade HCl for 1 day, followed by 1 day in hot high-purity HCl, 1 day in hot high-purity HNO_3, and 1 day in hot deionised water followed by rinsing in cold deionised water.

Preparation of candidate sulfide reference materials. Candidate sulfide reference materials were prepared from sulfide reference materials GBW07267, GBW07268 and GBW07270, purchased by the Institute of National Research Center for Geoanalysis, CAGS (Fig. 1). These certified reference materials are

Fig. 1 Photographs of the collected sulfide reference materials.

Table 1. The major and trace element compositions of three sulfide reference materials

RMs	Mineral	Major elements	Trace elements
GBW07267	pyrite	S, Fe	Ag, As, Bi, Cd, Co, Cu, Ga, Ge, Sb, Se, Sn, Te, Mn, Ni, Pb, Zn
GBW07268	chalcopyrite	S, Cu, Fe	Ag, As, Bi, Cd, Co, Ga, In, Sb, Se, Sn, Te, Mn, Ni, Pb, Zn
GBW07270	sphalerite	Zn, S, Fe	Ag, As, Bi, Cd, Co, Cu, Ga, Ge, In, Sb, Se, Sn, Te, Mn, Ni, Pb

are originally issued in small lots of ≈ 10 g per bottle. The major and trace element compositions of the sulfide reference materials are listed in Table 1, and the specific concentrations, directly retrieved from GeoReM - the Reference Material Database, are provided in Table S1. Approximately 300 g of each sulfide reference material was homogenised in a single FEP bottle by thorough shaking, then subdivided into fifty 6 g aliquots, each placed in a 10 mL polypropylene bottle and sealed for storage. These powders were used to test for isotopic homogeneity. The candidate pyrite (NWU-Fe), chalcopyrite (NWU-Cu) and sphalerite (NWU-Zn) reference materials were stored in an electronic drying box.

Digestion of sulfide reference materials. All chemical experiments were carried out at SKLCEEL. Approximately 5 mg of each sulfide reference material was weighed in the 15 mL pre-cleaned PFA vials. Each sulfide reference material was dissolved in 0.5 mL of concentrated HCl-HNO_3 (3:1, v/v) mixture twice at 80 °C for 12 h in a capped vial to ensure full dissolution. All dissolved samples were evaporated to dryness. Then all dried samples were re-dissolved in concentrated HNO_3 two times to ensure that the medium was completely converted to HNO_3 and evaporated to dryness again. Finally, all samples were individually dissolved in 3 mL of 2% HNO_3 (v/v) for the following experiment.

Chemical separation. Some samples were re-dissolved for the subsequent ion-exchange separation to obtain reference values to evaluate their suitability as a reference material using the without column chromatography method. For NWU-Fe, the column chromatography procedure followed was described by Chen *et al.* (2022)[30]. The dissolved samples were evaporated to dryness. Then all dried samples were redissolved with 1 mL of 6 mol L^{-1} HCl and 50 µL of H_2O_2, and they were subsequently evaporated to dryness again. This was done to eradicate possible organics and ensure that Fe was present as ferric iron in the solution. Finally, all samples were individually dissolved in 0.5 mL of 6 mol L^{-1} HCl + 0.01% H_2O_2 for chemical chromatography procedures. The resin column was cleaned alternatively with deionised water, 8 mol L^{-1} HNO_3, deionised water, 0.4 mol L^{-1} HCl and deionised water two times. After conditioning with 1 mL of 6 mol L^{-1} HCl + 0.01% H_2O_2, the dissolved sample containing 20 µg Fe was loaded into the column followed by loading 4 mL of 6 mol L^{-1} HCl + 0.01% H_2O_2 to elute most matrix elements. Fe fraction was eluted and collected with 5.5 mL of 0.4 mol L^{-1} HCl + 0.01% H_2O_2. The collected Fe fractions were dried up and dissolved in concentrated HNO_3 and H_2O_2 to remove possible organic matter during ion exchange column chromatographic separation. Then the dissolved solutions were re-evaporated to dryness. For NWU-Cu, the column chromatography procedure followed was described by Liu *et al.* (2014) and Zhang *et al.* (2020)[14, 29]. After cleaning the resin, 12mL of 8 mol L^{-1} HCl + 0.001% H_2O_2 was added to the column for conditioning. Then the dissolved sample containing about 20 µg Cu was loaded into the column followed by loading 8 mL of 8 mol L^{-1} HCl + 0.001% H_2O_2 to elute most matrix elements. Cu was collected in 22 mL of 8 mol L^{-1} HCl +0.001% H_2O_2. The collected Cu fractions were evaporated to dryness and dissolved in concentrated HNO_3. Then the dissolved solutions were re-evaporated to dryness. For NWU-Zn, the column chromatography procedure was described in Chen *et al.* (2016) and Nie *et al.* (2021)[31, 39]. The dissolved samples were evaporated to dryness, and then all dried samples were redissolved with 6 mol L^{-1} HCl for chemical chromatography procedures. After washing the resin with 2 mL of 0.5 mol L^{-1} HNO_3 and 2 mL of 6 mol L^{-1} HCl, 10 mL of 6 mol L^{-1} HCl was then added to the column for conditioning. The dissolved sample containing 20 µg of Zn was loaded, followed by loading 4 mL of 6 mol L^{-1} HCl to elute most matrix elements. Subsequently, the Co, Cu, Ga, and Fe were eluted using 6mL of 0.5 mol L^{-1} HCl. Zn was collected in 10 mL of 0.5 mol L^{-1} HNO_3. The collected Zn fractions were evaporated to dryness and dissolved in concentrated HNO_3. Then the dissolved solutions were re-evaporated to dryness. Before Fe-Cu-Zn isotope analysis, the samples were dissolved with 2% HNO_3 (v/v).

The total procedural blanks (from sample dissolution to mass spectrometry) were 5 ng for Fe, 2 ng for Cu, and 8 ng for Zn, which are negligible compared with 20 µg in samples loaded into the columns. Recoveries of Fe, Cu, and Zn were 99.5%, 99.8%, and 99.3%, respectively.

Mass spectrometry. Iron, Cu, and Zn isotopic compositions of sulfide reference materials were determined by using a double-focusing Neptune Plus MC-ICP-MS (Thermo Fisher Scientific, Bremen, Germany) at SKLCEEL. This instrument is equipped with eleven Faraday cups and five discrete dynode multipliers. The RF power was 1300 W. A 'wet' plasma with a standard sample cone, an H skimmer cone, a cyclone/double-pass spray chamber, and a low-flow PFA nebuliser (50 µL/min) was used to determine

Table 2. Results of homogeneity testing of between-bottle for the Fe-Cu-Zn isotopic ratios of NWU-Fe, NWU-Cu and NWU-Zn

Bottle No.	δ^{56}Fe			δ^{65}Cu			δ^{66}Zn		
	X_{i1}	X_{i2}	X_i	X_{i1}	X_{i2}	X_i	X_{i1}	X_{i2}	X_i
3	-0.35	-0.37	-0.36	0.44	0.40	0.42	-0.06	-0.05	-0.06
8	-0.33	-0.34	-0.33	0.45	0.41	0.43	-0.06	-0.06	-0.06
9	-0.37	-0.34	-0.35	0.44	0.41	0.42	-0.06	-0.06	-0.06
12	-0.33	-0.33	-0.33	0.44	0.43	0.44	-0.06	-0.06	-0.06
15	-0.34	-0.36	-0.35	0.43	0.43	0.43	-0.05	-0.06	-0.05
19	-0.33	-0.34	-0.35	0.43	0.41	0.42	-0.07	-0.06	-0.06
22	-0.35	-0.35	-0.35	0.44	0.43	0.44	-0.05	-0.05	-0.05
26	-0.33	-0.36	-0.34	0.43	0.45	0.44	-0.05	-0.06	-0.05
30	-0.34	-0.34	-0.34	0.45	0.44	0.45	-0.06	-0.06	-0.06
34	-0.35	-0.35	-0.35	0.45	0.42	0.43	-0.05	-0.06	-0.06
38	-0.37	-0.34	-0.36	0.44	0.42	0.43	-0.05	-0.07	-0.06
42	-0.36	-0.37	-0.37	0.41	0.39	0.40	-0.06	-0.06	-0.06
44	-0.35	-0.33	-0.34	0.40	0.39	0.39	-0.07	-0.06	-0.06
46	-0.35	-0.37	-0.36	0.41	0.39	0.40	-0.06	-0.05	-0.06
50	-0.38	-0.36	-0.37	0.40	0.42	0.41	-0.05	-0.04	-0.05

Table 3. ANOVA statistics for homogeneity testing of Fe-Cu-Zn isotopic ratios of NWU-Fe, NWU-Cu and NWU-Zn

	δ^{56}Fe	δ^{65}Cu	δ^{66}Zn
n		2	
m		15	
SS_{among}	0.0047	0.0070	0.0008
SS_{within}	0.0035	0.0041	0.0007
v_{among}		14	
v_{within}		15	
F	1.95	1.82	1.28
$F_{critical}$		2.42	

Fe-Cu-Zn isotopic ratios. During Fe isotope analysis, L2, L1, C, H1, H3 and H4 Faraday cups equipped with 10^{11} Ω amplifier resistors were used to collect ^{52}Cr, ^{53}Cr, ^{54}Fe+^{54}Cr, ^{56}Fe, ^{57}Fe and ^{58}Fe+^{58}Ni, respectively. In order to separate the Fe ions from the molecular interfering ions, the mass resolution was set at medium resolution (m/Δm = ~5000), and the measurement position was set at the center of the shoulder plateau on the Fe mass spectrum peak. The observed ^{53}Cr/^{52}Cr ratios (^{53}Cr/^{52}Cr = 0.113887) were used for calculating the mass bias factor with the exponential law, and ^{52}Cr (^{54}Cr/^{52}Cr = 0.028222) and ^{53}Cr (^{54}Cr/^{53}Cr = 0.247807) were monitored to subtract the interference of ^{54}Cr on ^{54}Fe isotopes with the exponential law, respectively. The standard-sample bracketing (SSB) method was selected to correct the instrumental mass bias. Iron isotope results were firstly expressed as a per mil deviation relative to the in-house standard NWU-Fe and then converted into values relative to the reference material IRMM-014. During the Cu isotope analysis, L4, L2, H2 and H4 Faraday cups were used to collect ^{63}Cu, ^{65}Cu, ^{69}Ga and ^{71}Ga, respectively. The SSB method combined with NIST SRM 994 Ga (^{69}Ga/^{71}Ga = 1.50676) as an internal reference material was selected to correct the instrumental mass bias. The in-house standard NWU-Cu-B served as a bracketing standard. Copper isotope results were firstly expressed as a per mil deviation relative to the NWU-Cu-B and then converted into values relative to the reference material NIST SRM 976. During Zn isotope analysis, L3, L2, L1, C, H1, H2, and H3 Faraday cups were used to collect ^{62}Ni, ^{63}Cu, ^{64}Zn, ^{65}Cu, ^{66}Zn, ^{67}Zn, and ^{68}Zn, respectively. Zinc isotope ratios were measured in medium-resolution mode to resolve potential polyatomic interferences. The SSB method combined with NWU-Cu-B (^{65}Cu/^{63}Cu = 0.44689) as an internal reference material was selected to correct the instrumental mass bias. The in-house standard solution NIST SRM 683 served as a bracketing calibrator. Zinc isotope results were firstly expressed as a per mil deviation relative to the NIST SRM 683 and then converted into values relative to the reference material JMC-Lyon.

Iron, Cu and Zn isotopic values relative to IRMM-014, NIST SRM 976 and JMC-Lyon, respectively, were calculated as follows:

$$\delta^{56}Fe = ((^{56}Fe/^{54}Fe)_{sample} / (^{56}Fe/^{54}Fe)_{IRMM014} - 1)*1000 \quad (1)$$

$$\delta^{65}Cu = ((^{65}Cu/^{63}Cu)_{sample} / (^{65}Cu/^{63}Cu)_{NIST\ SRM976} - 1)*1000 \quad (2)$$

$$\delta^{66}Zn = ((^{66}Zn/^{64}Zn)_{sample} / (^{66}Zn/^{64}Zn)_{JMC\text{-}Lyon} - 1)*1000 \quad (3)$$

The detailed mass spectrometry analyses for Fe, Cu and Zn can be found in Chen *et al*. (2022), Zhang *et al*. (2020) and Nie *et al*. (2022), respectively[29-31].

RESULTS AND DISCUSSION

Homogeneity of sulfide reference materials. All isotopic measurements for the homogeneity assessment were performed using MC-ICP-MS without column chemistry. Homogeneity is a critical requirement for reference materials and was ensured in this study by adhering to international regulations for reference material preparation (ISO Guide 35 2006). Isotopic data for the homogeneity test of sulfide powders were collected under strict repeatability conditions, ensuring consistency across measurements. All analyses were conducted in the SKLCEEL using

Table 4. Long-term stability results of Fe, Cu and Zn isotopic compositions over 36 months

Time (month)	δ^{56}Fe	2s	*n* [a]	Time (month)	δ^{65}Cu	2s	*n* [a]	Time (month)	δ^{66}Zn	2s	*n* [a]
0	-0.31	0.06	8	0	0.42	0.04	49	0	-0.06	0.03	40
2	-0.34	0.04	12	1	0.43	0.02	41	2	-0.05	0.03	20
5	-0.36	0.04	41	3	0.44	0.04	36	5	-0.05	0.03	32
7	-0.33	0.05	9	5	0.46	0.02	12	7	-0.06	0.02	120
9	-0.35	0.04	90	6	0.45	0.03	10	8	-0.03	0.03	8
12	-0.37	0.04	14	9	0.45	0.01	12	11	-0.02	0.03	10
15	-0.33	0.05	5	11	0.44	0.03	11	13	-0.05	0.02	20
17	-0.35	0.03	6	12	0.43	0.03	19	14	-0.02	0.03	6
20	-0.31	0.06	3	14	0.43	0.04	24	16	-0.05	0.02	24
22	-0.36	0.04	33	15	0.42	0.04	120	18	-0.02	0.04	6
24	-0.35	0.07	21	17	0.41	0.05	48	20	-0.04	0.02	8
26	-0.36	0.04	30	20	0.39	0.02	24	22	-0.04	0.03	8
28	-0.35	0.05	12	23	0.43	0.03	35	24	-0.06	0.01	20
30	-0.36	0.03	13	24	0.39	0.01	5	26	-0.03	0.01	4
31	-0.37	0.03	10	25	0.43	0.03	6	27	-0.07	0.02	20
33	-0.35	0.06	30	27	0.45	0.02	8	29	-0.03	0.02	8
34	-0.35	0.07	22	29	0.45	0.02	13	30	-0.03	0.02	12
36	-0.35	0.06	15	30	0.45	0.02	12	32	-0.07	0.02	22
				31	0.41	0.03	11	33	-0.05	0.01	4
				32	0.39	0.04	12	34	-0.08	0.02	12
				34	0.41	0.03	28	36	-0.02	0.02	44
				36	0.44	0.03	44				
Mean	**-0.35**	**0.03**	**374**	Mean	**0.43**	**0.04**	**485**	Mean	**-0.04**	**0.03**	**388**

[a] Number of repeat measurements of these reference materials by MC-ICP-MS.

the same instrument, method, and analyst[40, 41]. Fifteen bottles of each reference material (NWU-Fe, NWU-Cu, and NWU-Zn) were randomly selected for homogeneity evaluation. Two subsamples were taken from each bottle and treated as independent samples. Each sample was analysed at least three to four times to obtain a single δ value. Results of replicate isotopic analyses are represented as X_{i1} and X_{i2} in Table 2, and the average value of duplicate pairs is expressed as X_i. The between-bottle inhomogeneity was calculated using a single-factor ANOVA test. The experimental F ratio is defined as the ratio of among-bottle variance (S^2_{among}) to within-bottle variance (S^2_{within}):

$$F = \frac{S^2_{among}}{S^2_{within}} \tag{4}$$

where S^2_{among} is the ratio of among-bottle sums of squares (SS_{among}) to the associated degrees of freedom (v_{among}), and S^2_{within} is the ratio of within-bottle sums of squares (SS_{within}) to the associated degrees of freedom (v_{within}):

$$S^2_{among} = SS_{among}/v_{among} \tag{5}$$

$$S^2_{within} = SS_{within}/v_{within} \tag{6}$$

where v_{among} and v_{within} depend on the number of bottles from which samples are taken (m) and the number of replicate measurements for each bottle (n), the degrees of freedom v_{among} and v_{within} are calculated as follows:

$$v_{among} = m - 1 \tag{7}$$

$$v_{within} = m \times n - m \tag{8}$$

Isotopic ratios of between-bottle and ANOVA tests for Fe, Cu, and Zn are presented in Tables 2 and 3, respectively. F-testing indicates that the homogeneity testing results were insignificant 5%)[40-43], demonstrating that the sulfide powders exhibit excellent homogeneity at the 95% confidence level. Detailed measurement results are provided in Appendix S1.

Stability of sulfide reference materials. Stability monitoring was also carried out without column chromatography. The long-term stability of NWU-Fe, NWU-Cu, and NWU-Zn was evaluated by monitoring δ^{56}Fe, δ^{65}Cu, and δ^{66}Zn values over 36 months (Fig. 2; Table 4), and the average values of δ^{56}Fe, δ^{65}Cu, and δ^{66}Zn are -0.35 ± 0.03‰ (2s, n = 374), 0.43 ± 0.04‰ (2s, n = 485), and -0.04

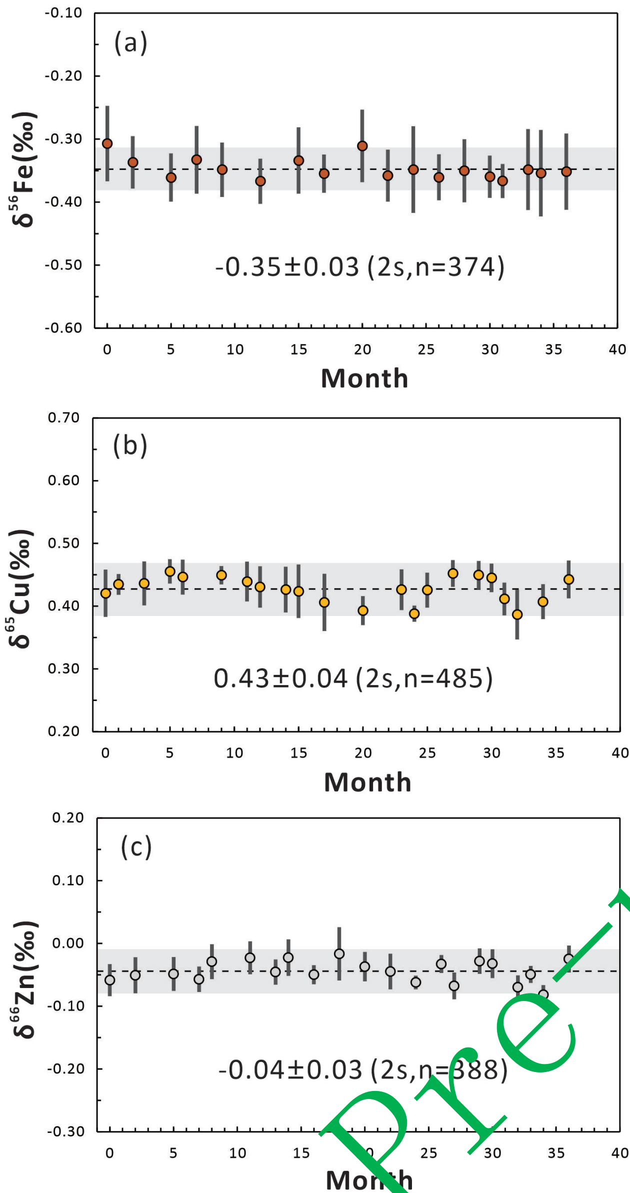


Fig. 2 Long-term stability results of Fe, Cu and Zn isotopic compositions of (a) NWU-Fe, (b) NWU-Cu and (c) NWU-Zn. Bars represent 2s.

Table 5. Results of stability tests for NWU-Fe, NWU-Cu and NWU-Zn

NWU	$\delta^{56}Fe$	$\delta^{65}Cu$	$\delta^{66}Zn$
b_1	-0.00065	-0.00053	-0.00015
b_0	-0.33449	0.43694	-0.04124
s^2	0.00025	0.00042	0.00033
s	0.01579	0.02045	0.01815
$s_{(b1)}$	0.00033	0.00039	0.00036
t	36	36	36
$t_{0.95,n-2}$	2.599	2.606	1.588
$t_{0.95,n-2}\cdot s_{(b1)}$	0.00086	0.00102	0.00058

± 0.03‰ (2s, n = 388), respectively. Individual MC-ICP-MS analytical results can be found in Appendix S2. To assess stability, a linear regression model was employed, as recommended by the ISO Guide 35 (2006), expressed as:

$$Y = b_0 + b_1 X + \varepsilon \qquad (9)$$

Here, b_0 and b_1 represent regression coefficients, b_0 is the intercept of the regression line, b_1 is the slope of the regression line, and ε denotes the random error term. X represents the independent variable (time in months), and Y represents the dependent variable (e.g., $\delta^{56}Fe$, $\delta^{65}Cu$, or $\delta^{66}Zn$ values). By conducting regression analysis on the data, the values of b_0 and b_1 can be calculated, thereby determining the linear relationship between Y and X. The slope of the regression line was calculated using:

$$b_1 = \frac{\sum_{i=1}^{n}(X_i-\bar{X})(Y_i-\bar{Y})}{\sum_{i=1}^{n}(X_i-\bar{X})^2} \qquad (10)$$

and the intercept was determined by:

$$b_0 = \bar{Y} - b_1\bar{X} \qquad (11)$$

The standard deviation of a point along the line can be computed from s^2 and $s(b_1)$ as follows:

$$s^2 = \frac{\sum_{i=1}^{n}(Y_i-b_0-b_1X_i)^2}{n-2} \qquad (12)$$

$$s(b_1) = \frac{s}{\sqrt{\sum_{i=1}^{n}(X_i-\bar{X})^2}} \qquad (13)$$

Using Equation (10) and the appropriate Student's t-factor for (n − 2) degrees of freedom at a 95% confidence level, the critical t-values were calculated as $t_{0.95,(n-2)} = 2.599$ for Fe in NWU-Fe, 2.606 for Cu in NWU-Cu, and 1.588 for Zn in NWU-Zn. A statistical test confirmed that $|b_1| < t_{0.95,(n-2)} \cdot s(b_1)$, indicating that the slope was not significantly different from zero. The detailed calculation process can be found in Appendix S3. This result demonstrates that the $\delta^{56}Fe$, $\delta^{65}Cu$ and $\delta^{66}Zn$ values for NWU-Fe, NWU-Cu, and NWU-Zn exhibited no statistically significant instability over the three-year monitoring period (Table 5). The findings validate the suitability of these reference materials for long-term isotopic measurements. Additionally, the results from NWU-Fe, NWU-Cu, and NWU-Zn further confirm their homogeneity and reliability, demonstrating great potential as reference materials.

Fe, Cu and Zn isotopic compositions of NWU-Fe, NWU-Cu and NWU-Zn. Interlaboratory comparisons among the State Key Laboratory of Continental Evolution and Early Life at Northwest University (NWU), the State Key Laboratory of Lithospheric and Environmental Coevolution at Institute of Geology and Geophysics, Chinese Academy of Sciences (IGGCAS), and the State Key Laboratory for Nuclear Resources and Environment at

Table 6. Iron, Cu and Zn isotopic compositions of NWU-Fe, NWU-Cu and NWU-Zn sulfide powder reference materials, as determined by participating laboratories

Institution/Laboratory	Replicates	$\delta^{56}Fe \pm 2s$	$\delta^{65}Cu \pm 2s$	$\delta^{66}Zn \pm 2s$
State Key Laboratory of Continental Evolution and Early Life (NWU)	1	-0.35 ± 0.03	0.44 ± 0.02	-0.02 ± 0.03
	2	-0.37 ± 0.06	0.45 ± 0.02	-0.03 ± 0.02
	3	-0.36 ± 0.02	0.44 ± 0.01	-0.03 ± 0.01
	4	-0.37 ± 0.02	0.44 ± 0.01	-0.02 ± 0.03
	Mean ± 2s	-0.36 ± 0.02	0.44 ± 0.01	-0.02 ± 0.02
State Key Laboratory of Lithospheric and Environmental Coevolution (IGGCAS)	1	-0.40 ± 0.03	0.39 ± 0.04	-0.05 ± 0.03
	2	-0.40 ± 0.03	0.38 ± 0.02	-0.08 ± 0.02
	3	-0.41 ± 0.02	0.39 ± 0.03	-0.04 ± 0.02
	4	-0.39 ± 0.02	0.40 ± 0.02	-0.05 ± 0.07
	Mean ± 2s	-0.40 ± 0.01	0.39 ± 0.02	-0.05± 0.03
State Key Laboratory for Nuclear Resources and Environment (ECUT)	1	-0.37 ± 0.03	0.48 ± 0.02	-0.02 ± 0.02
	2	-0.37 ± 0.01	0.47 ± 0.02	-0.02 ± 0.02
	3	-0.37 ± 0.03	0.48 ± 0.02	-0.03 ± 0.02
	4	-0.36 ± 0.01	0.48 ± 0.02	-0.03 ± 0.02
	Mean ± 2s	-0.37 ± 0.01	0.48 ± 0.01	-0.03 ± 0.01
Grand mean ± 2s		-0.38 ± 0.02	0.44 ± 0.04	-0.03 ± 0.02

Note: Each laboratory received four aliquots for each reference material (NWU-Fe, NWU-Cu, NWU-Zn) and performed at least four replicate analyses per aliquot. Results are expressed as mean ± 2s. All measurements were performed using MC-ICP-MS analysis without column chromatography.

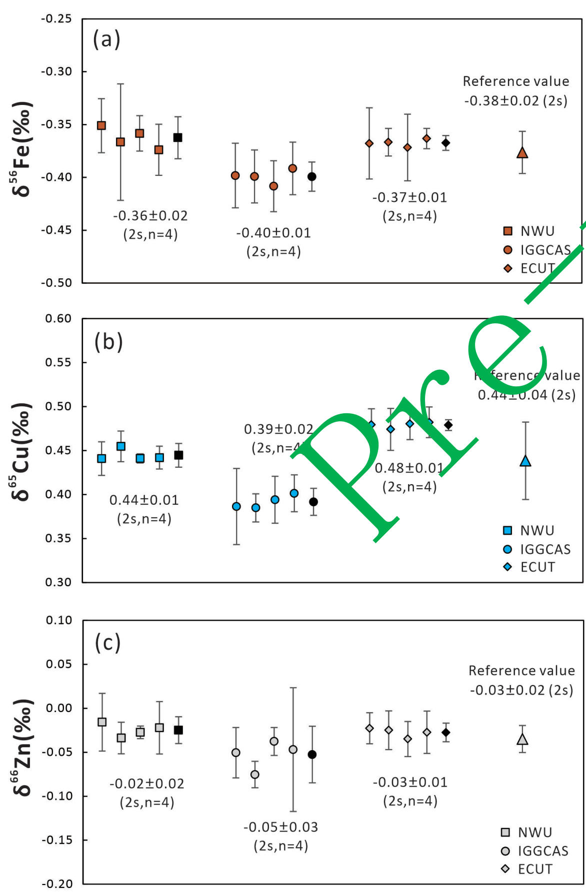


Fig. 3 Inter-laboratory comparison of (a) NWU-Fe, (b) NWU-Cu and (c) NWU-Zn isotope compositions. Black solid dots indicate the mean of the four bottles, error bars are 2s. Reference values were established as the unweighted means of inter-laboratory results from three independent laboratories.

East China University of Technology (ECUT) demonstrated harmonised results for the reference isotopic compositions, despite variations in analytical methodologies. All participating laboratories performed MC-ICP-MS without column chromatography, following the same direct analysis protocol. The resulting isotope ratios are consistent with the reference values obtained with column chemistry, confirming the accuracy of the without column chromatography method[29-31]. For intercomparison, four aliquots of each reference material (NWU-Fe, NWU-Cu, NWU-Zn) were distributed to three laboratories (NWU, IGGCAS, ECUT). Each laboratory analysed every aliquot at least four times by MC-ICP-MS without column chromatography. Results (Table 6; Fig. 3) are reported as means ± 2s, with reference values derived from the unweighted means of interlaboratory data. Individual MC-ICP-MS analytical results can be found in Appendix S4.

Figure 3 shows that there are slight systematic differences between the Fe, Cu, and Zn isotopic data provided by IGGCAS and those from NWU and ECUT. These differences may be attributed to two primary factors. First, IGGCAS used a Nu Sapphire MC-ICP-MS operating in low mass resolution mode ($m/\Delta m \approx 300$), whereas NWU and ECUT employed the Neptune Plus in medium mass resolution mode ($m/\Delta m \approx 5000$). The Nu Sapphire and Neptune Plus instruments have different designs and operational parameters, which can lead to minor variations in isotopic fractionation and interference correction. Additionally, the lower mass resolution of the Nu Sapphire instrument may result in slightly less precise separation of isotopes and potential interferences, contributing to the observed differences. Second, the sample introduction systems differ between the laboratories. IGGCAS employed a 100 µL/min PFA nebuliser, whereas NWU and ECUT used 50 µL/min PFA nebulisers. These differences in nebuliser flow rates can result in slight changes in sample transmission efficiency and nebulisation characteristics, potentially contributing to the observed isotopic variations. Overall, these differences are within the acceptable uncertainties of the measurements and do not compromise the certified isotopic compositions. The high consistency of inter-laboratory results validates the analytical precision of participating institutions in measuring Fe, Cu, and Zn isotopic ratios. The certified isotopic compositions of these reference materials, derived from the unweighted means of inter-laboratory results, are as follows: NWU-Fe $\delta^{56}Fe = -0.38 \pm 0.03‰$; NWU-Cu $\delta^{65}Cu = 0.44 \pm 0.04‰$; and NWU-Zn $\delta^{66}Zn = -0.04 \pm 0.02‰$.

CONCLUSION

Three sulfide reference materials, NWU-Fe, NWU-Cu and NWU-Zn, have been developed for Fe, Cu and Zn isotope analyses without column chromatography. These materials are specifically designed to validate and monitor direct measurement methods for simple-matrix minerals, thereby enhancing the accuracy and reliability of Fe, Cu, and Zn isotopic analyses in such minerals. Their isotopic compositions were determined by multiple laboratories using MC-ICP-MS. These materials exhibit appropriate offset values from zero-delta, along with excellent homogeneity and stability, making them well-suited for use in instrument calibration and analyst training.

All these reference materials are currently available in large amounts for sharing and can be requested by laboratories worldwide by contacting Honglin Yuan at the SKLCEEL (e-mail: yhlsklcd@126.com).

ASSOCIATED CONTENT

The supporting information (Table S1 and Appendix S1-S4) is available at https://www.at-spectrosc.com.

AUTHOR INFORMATION

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Notes

The authors declare no competing financial interest.

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REFERENCES

1. D. C. M. Fox, L. E. McGee, J. Farkaš, J. L. Payne, S. C. Spinks, M. Barham, and M. Aspandiar, *Geochem. Geophys. Geosyst.*, 2023, **24**, e2023GC011019. https://doi.org/10.1029/2023GC011019
2. B. Mahan, R. Mathur, I. Sanislav, P. Rea, and P. Dirks, *Appl. Geochem.*, 2023, **148**, 105519. https://doi.org/10.1016/j.apgeochem.2022.105519
3. Y. Wang, X. K. Zhu, C. Tang, J. W. Mao, and Z. S. Chang, *Ore Geol. Rev.*, 2021, **130**, 103946. https://doi.org/10.1016/j.oregeorev.2020.103946
4. C. W. Zhu, J. Wang, J. W. Zhang, X. C. Chen, H. F. Fan, Y. X. Zhang, T. Yang, and H. J. Wen, *Ore Geol. Rev.*, 2020, **117**, 103274. https://doi.org/10.1016/j.oregeorev.2019.103274
5. Y. Tang, G. L. Han, L. Man, J. Zeng, and R. Qu, *Sci. Total Environ.*, 2023, **878**, 162964. https://doi.org/10.1016/j.scitotenv.2023.162964
6. D.-J. Joe, M.-S. Choi, J.-H. Lee, C.-K. Kim, M.-S. Choi, and H.-S. Shin, *Environ. Sci. Pollut. Res.*, 2024, **31**, 20521-20533. https://doi.org/10.1007/s11356-024-32508-6
7. Y. Jiang, J. T. Kang, S. Y. Liao, S. M. Elardo, K. Q. Zong, S. J. Wang, C. Nie, P. Y. Li, Z. J. Yin, F. Huang, and W. B. Hsu, *Astrophys. J. Lett.*, 2023, **945**, L26. https://doi.org/10.3847/2041-8213/acbd31
8. M.-N. Decraene, J. Marin-Carbonne, C. Thomazo, A. Brayard, A.-S. Bouvier, B. Bomou, T. Adatte, and N. Olivier, *Palaeogeogr. Palaeoclimatol. Palaeoecol.*, 2023, **617**, 111507. https://doi.org/10.1016/j.palaeo.2023.111507

9. J. A. Stephens, M. N. Ducea, D. J. Killick, and J. Ruiz, *J. Archaeol. Sci.*, 2021, **127**, 105334. https://doi.org/10.1016/j.jas.2021.105334
10. F. Moynier, A. Merland, D. Rigoussen, J. Moureau, M. Paquet, B. Mahan, and M. Le Borgne, *Metallomics*, 2022, **14**, mfac017. https://doi.org/10.1093/mtomcs/mfac017
11. K. V. Sullivan, R. E. T. Moore, and F. Vanhaecke, *Metallomics*, 2023, **15**(3), mfad012. https://doi.org/10.1093/mtomcs/mfad012
12. C. N. Maréchal, P. Télouk, and F. Albarède, *Chem. Geol.*, 1999, **156**, 251-273. https://doi.org/10.1016/S0009-2541(98)00191-0
13. C. Maréchal and F. Albarède, *Geochim. Cosmochim. Acta*, 2002, **66**, 1499-1509. https://doi.org/10.1016/S0016-7037(01)00815-8
14. S. -A. Liu, D. D. Li, S. G. Li, F. Z. Teng, S. Ke, Y. S. He, and Y. H. Lu, *J. Anal. At. Spectrom.*, 2014, **29**, 122-133. https://doi.org/10.1039/c3ja50232e
15. Y. S. He, S. Ke, F.-Z. Teng, T. T. Wang, H. J. Wu, Y. H. Lu, and S. G. Li, *Geostand. Geoanal. Res.*, 2015, **39**, 341-356. https://doi.org/10.1111/j.1751-908X.2014.00304.x
16. K. Kehm, E. H. Hauri, C. M. O' D. Alexander, and R. W. Carlson, *Geochim. Cosmochim. Acta*, 2003, **67**, 2879-2891. https://doi.org/10.1016/S0016-7037(03)00080-2
17. G. H. Zhu, J. L. Ma, G. J. Wei, and Y. J. An, *J. Anal. At. Spectrom.*, 2020, **35**, 873-877. https://doi.org/10.1039/c9ja00446g
18. P. A. Sossi, G. P. Halverson, O. Nebel, and S. M. Eggins, *Geostand. Geoanal. Res.*, 2015, **39**, 129-149. https://doi.org/10.1111/j.1751-908X.2014.00298.x
19. Z. A. Bao, N. Lv, K. Y. Chen, Y. Luan, X. H. Sun, C. L. Zong, and H. L. Yuan, *Geostand. Geoanal. Res.*, 2021, **45**, 401-418. https://doi.org/10.1111/ggr.12372
20. Z. A. Bao, X. J. Nie, K. Y. Chen, C. L. Zong, N. Lv, and H. L. Yuan, *Geostand. Geoanal. Res.*, 2022, **46**, 433-449. https://doi.org/10.1111/ggr.12427
21. L. Chen, Y. Liu, Y. Li, Q. L. Li, and X. H. Li, *J. Anal. At. Spectrom.*, 2021, **36**, 1431-1440. https://doi.org/10.1039/d1ja00029b
22. K. Y. Chen, H. L. Yuan, Z. A. Bao, and N. Lv, *Atom. Spectrosc.*, 2021, **42**, 282-293. https://doi.org/10.46770/AS.2021.703
23. K. Ikehata, K. Notsu, and T. Hirata, *J. Anal. At. Spectrom.*, 2008, **23**, 1003-1008. https://doi.org/10.1039/b801044g
24. M. Lazarov and I. Horn, *Spectrochim. Acta Part B*, 2015, **111**, 64-73. https://doi.org/10.1016/j.sab.2015.06.013
25. L. Xu, W. Zhang, T. Luo, J. H. Yang, and Z. C. Hu, *J. Anal. At. Spectrom.*, 2021, **36**, 747-757. https://doi.org/10.1039/d0ja00465k
26. Q. C. Yang, H. M. Gong, X. H. Wang, M. Duan, Y. Y. Xiao, and W. D. Sun, *J. Anal. At. Spectrom.*, 2022, **37**, 1000-1009. https://doi.org/10.1039/d1ja00410g
27. X. Y. Zheng, B. L. Beard, and C. M. Johnson, *J. Anal. At. Spectrom.*, 2018, **33**, 68-83. https://doi.org/10.1039/c7ja00272f
28. N. Lv, Z. A. Bao, K. Y. Chen, K. Wu, and H. L. Yuan, *Sci. China Earth Sci.*, 2022, **65**, 2005-2017. https://doi.org/10.1007/s11430-021-9943-y
29. Y. Zhang, Z. A. Bao, N. Lv, K. Y. Chen, C. L. Zong, and H. L. Yuan, *Front. Chem.*, 2020, **8**, 1-11. https://doi.org/10.3389/fchem.2020.00609
30. K. Y. Chen, Z. A. Bao, H. L. Yuan, and N. Lv, *J. Anal. At. Spectrom.*, 2022, **37**, 249-263. https://doi.org/10.1039/d1ja00381j
31. X. J. Nie, Z. A. Bao, P. Liang, K. Y. Chen, C. L. Zong, and H. L. Yuan, *Atom. Spectrosc.*, 2022, **43**, 117-125. https://doi.org/10.46770/AS.2021.910
32. J.-F. He, G.-G. Wang, M. J. Brzozowski, P. Ni, X.-L. Luo, J. Mu, and W. Q. Li, *Miner. Deposita*, 2025, **60**, 811-831. https://doi.org/10.1007/s00126-024-01321-z
33. X. K. Zhu, R. K. O'Nions, Y. Guo, N. S. Belshaw, and D. Rickard, *Chem. Geol.*, 2000, **163**, 139-149. https://doi.org/10.1016/S0009-2541(99)00076-5
34. R. Mathur, J. Ruiz, S. Titley, L. Liermann, H. Buss, and S. Brantley, *Geochim. Cosmochim. Acta*, 2005, **69**, 5233-5246. https://doi.org/10.1016/j.gca.2005.06.022
35. R. Mathur and Y. Zhao, in: Huston, D., Gutzmer, J. (Eds.), Isotopes in Economic Geology, Metallogenesis and Exploration, Springer Nature, 2023, pp433-450. https://doi.org/10.1007/978-3-031-27897-6_14
36. J. Majzlan, R. Mathur, R. Milovský, and S. Milovská, *Mineral. Mag.*, 2022, **86**, 644-651. https://doi.org/10.1180/mgm.2021.77
37. S. Graham, N. Pearson, S. Jackson, W. Griffin, and S. Y. O'Reilly, *Chem. Geol.*, 2004, **207**, 147-169. https://doi.org/10.1016/j.chemgeo.2004.02.009
38. N. S. Belshaw, X. K. Zhu, Y. Guo, and R. K. O'Nions, *Int. J. Mass Spectrom.*, 2000, **197**, 191-195. https://doi.org/10.1016/S1387-3806(99)00245-6
39. S. Chen, Y. C. Liu, J. Y. Hu, Z. F. Zhang, Z. H. Hou, F. Huang, and H. M. Yu, *Geostand. Geoanal. Res.*, 2016, **40**, 417-432. https://doi.org/10.1111/j.1751-908X.2015.00377.x
40. J. S. Kane, P. J. Potts, M. Wiedenbeck, J. Carignan, and S. Wilson, *Geostand. Newsl.*, 2003, **27**, 227-244. https://doi.org/10.1111/j.1751-908X.2003.tb00724.x
41. J. Li, S.-h. Tang, X.-k. Zhu, Z.-h. Li, S.-Z. Li, B. Yan, Y. Wang, J. Sun, Y. Shi, A. G. Dong, N. S. Belshaw, X. C. Zhang, S.-a. Liu, J.-h. Liu, D. Wang, S.-y. Jiang, K. Hou, and A. S. Cohen, *Geostand. Geoanal. Res.*, 2019, **43**, 163-175. https://doi.org/10.1111/ggr.12241
42. N. Lv, Z. Bao, K. Y. Chen, C. L. Zong, Y. Zhang, and H. L. Yuan, *Geostand. Geoanal. Res.*, 2022, **46**, 451-463. https://doi.org/10.1111/ggr.12440
43. N. Lv, Z. Bao, X. J. Nie, K. Y. Chen, Y. Zhang, and H. L. Yuan, *Geostand. Geoanal. Res.*, 2024, **48**, 411-421. https://doi.org/10.1111/ggr.12544